60. (New) A hypermutable, non-human, transgenic mammal wherein at least 50% of the cells of said mammal comprise a dominant negative allele of a mismatch repair gene.

61. (New) A hypermutable, non-human, transgenic mammal produced by a process comprising introducing a polynucleotide comprising a sequence encoding a dominant negative allele of a mismatch repair gene into said mammal, whereby said mammal becomes hypermutable.

62. (New) A method of making a hypermutable, non-human, mammalian, fertilized egg comprising introducing into said mammalian fertilized egg a polynucleotide comprising a sequence encoding a dominant negative allele of a mismatch repair gene, whereby said mammalian fertilized egg becomes hypermutable.

Remarks

Applicants have added new claims 60-62. Support for these claims may be found, for example, in the original claims 28, 1, 12, 40, 45 and 50. The added claims provide antecedent basis for the amended, pending claims.

Applicants have amended claims 29, 52, 53 and 58, as helpfully suggested by the Examiner to include the feature that the transgenic mammal is "non-human." Newly added claims also recite "non-human" transgenic mammal. Claim 29 has been further amended to recite "mammal" rather than "animal" to provide consistency of claim language.

Claims 13, 14, 52, 53 and 58 have been amended to depend from the newly added claims which provide antecedent basis for the terms "the method of claim [12] 62," "the mismatch repair gene," "the hypermutable non-human transgenic animal of claim [28] 60," "the hypermutable non-human transgenic animal of claim [50] 61," and "the allele." Furthermore, Applicants have amended claims 18 and 19 to recite "said mismatch repair gene" rather than "allele" to provide consistency of claim language. No new matter has been added. It is respectfully submitted that the claims are in proper form for allowance.

We turn now to the merits of the invention. The Applicants have discovered a novel method of introducing mutations into the genomes of animals by introducing a dominant negative allele of a mismatch repair gene into the mammal to create hypermutable transgenic animals, and transgenic animals made by the method.

The Office Action alleges that the Specification does not provide adequate guidance as to making transgenic animals. The Office Action further alleges that the Specification merely provides a general statement that making transgenic animals is known in the art, and cursory statements regarding transgenic animals. Applicants respectfully disagree. The Specification as filed provides guidance as to making transgenic animals by a number of well-recognized methods in the art for preparing transgenic animals.

Microinjection of fertilized eggs

We invite the Examiner's attention to the Specification at page 9, lines 22-28, wherein it is described that one method of producing transgenic animals is to inject the polynucleotide into the fertilized egg of a donor animal, which is produced *in vitro* using sperm and eggs of donor

animals. The injected, fertilized eggs are then placed in a pseudo-pregnant animal of the same species. The passage further indicates that the pseudo-pregnant animal is produced by hormone treatments.

Microinjection or Transfection of Embryonic Stem Cells

The Specification also describes an alternative method of making transgenic animals involving injection or transfection of embryonic stem cells at page 9, lines 29-31 through page 10, lines 1-4. In this passage the Specification describes that the polynucleotide is either injected into the embryonic stem cell, or transfected into the embryonic stem cell. Thereafter, the modified embryonic stem cells are re-introduced into the developing embryo. The Specification further teaches that in this method, the polynucleotide must be incorporated into the germline of the transgenic animal or the gene will not be passed on to the progeny. Therefore, as the Specification notes at page 10, lines 2-4, that the animals produced by this method should be tested to determine whether the transgene has been introduced into the germline of the animal.

The Office Action alleges that the Specification does not provide sufficient guidance in making and using transgenic animals without undue experimentation. Applicants respectfully traverse. The Specification provides a description of at least two methods for producing transgenic animals. The level of skill of one of ordinary skill in the art is high, and armed with the disclosure and knowledge of the prior art, the skilled artisan would be able to make and use the claimed transgenic animals without undue experimentation. The Federal Circuit noted in *In re Buchner* that everything necessary to practice the invention need not be disclosed. In fact, what is well-known is best omitted. *In re Buchner* 929 F.2d 660,661, 18 U.S.P.Q.2d 1331, 1332

(Fed. Cir. 1991). Thus, it is not required that the Applicants supply a detailed protocol for producing transgenic animals.

The Examiner describes the "current state of the transgenic animal research" as problematic with respect to certain animal species in that there are often longer gestation times, reduced litter sizes, the number of eggs needed to inject, low efficiency of gene integration and method of introduction of transgenes. Notably none of these so-called limitations demonstrate that transgenic animals are not enabled. Each of these "problems" merely addresses the drawbacks of producing transgenics of species other than mice. This does not show that making transgenics in other animals is not enabled. In fact, it is evidence that producing transgenic animals of various types is possible, albeit without the convenience of a mouse's shorter gestation period, larger litter sizes, and number of eggs obtainable to inject and implant (and ease of obtaining them). Low efficiency of integration is simply a compounding factor with the latter. We invite the Examiner's attention to Cameron (1997) "Recent advances in transgenic technology" *Mol. Biotechnol.* 7(3):253-65 (Abstract)(article also cited by the Examiner) which describes how "transgenic science has played an important part in increasing understanding of the complex processes that underlie gene regulation, and this in turn has assisted in the design of transgene constructs expressed in a tightly regulated and faithful manner." The Abstract also notes that the production of transgenic livestock "is an inefficient process compared to that of laboratory models, and the lack of totipotential embryonic stem (ES) cell lines in farm animal species hampers the development of this area of work. This article highlights recent progress in efficient transgene expression systems, and the current efforts being made to find alternative means of generating transgenic livestock." Thus, at the time of filing, it was recognized that

producing transgenic livestock was not as efficient as producing transgenic mice. Nonetheless the former was known at the time of filing and recent advances had been made in transgenic expression systems. Applicants note that in support of his argument, the Examiner cites Mullins *et al.* (1993) *Hypertension* 22:630-633. However, the subject Application was filed in 1998, five years after publication of Mullins *et al.* Applicants submit that Mullins *et al.* does not fairly represent the state of the art of transgenic animals at the time the Application was filed. The Office Action further cites Hammer *et al.* (1990) *Cell* 63:1099-1112 for the proposition that different effects of a transgene may be seen in different species. Again, 1990 hardly represents the "state of the art" at the time the subject Application was filed.

The Examiner expresses concerns regarding which promoters could be used for different species or what levels of gene expression would be required to produce a phenotype. It is well within the purview of the skilled artisan that species-specific promoters may be used in constructing a given transgenic construct. Also, optimization of the system is within the purview of the skilled artisan. A considerable amount of experimentation may be required to practice the invention, provided it is not undue (*see In re Angstadt*, 537 F.2d 498,504, 190 U.S.P.Q. 214, 219 (CCPA 1976)). The Examiner's reliance on references such as Seidel J. (1993) *Anim. Sci.* 71 (Suppl. 3):26-33, is misplaced since the references show that production of certain transgenic animals is not as efficient as production of transgenic mice. Longer gestational periods, longer times for animals to reach reproductive age, potential variability in expression from animal to animal, *etc.* are of no consequence in an enablement analysis. Applicants are not obliged to demonstrate that the execution of the methods will be without some inconvenience.

In short, while the Examiner raises the specter that production of transgenic animals in many species may be *inconvenient*, the Examiner fails to make a *prima facie* case that the claims are not *enabled*.

The claims encompass transgenic animals that comprise a *dominant negative* allele of a mismatch repair gene, such as a dominant negative allele of PMS2. Contrary to the Office Action's assertions, the Specification provides ample guidance as to the use of transgenic animals comprising a dominant negative allele of a mismatch repair gene. We invite the Examiner's attention to the Specification at page 10, lines 7-24, wherein it is taught that once a transgenic animal is produced, a gene of interest may be screened for mutations (as the transgenic animal is subject to hypermutation). The gene of interest may be screened at the genetic level by examining the sequence of cDNA, genomic DNA, or RNA, or the phenotype may be examined. Mutant phenotypes may be screened by detecting alterations in electrophoretic mobility, spectroscopic properties, and other physical and structural characteristics. One may also examine alterations in protein function *in situ*, in isolated form, or in model systems. In short, one may use the transgenic animals to examine properties of genes of interest which have been mutated through hypermutation.

The Specification Satisfies the Written Description Requirement

The Office Action alleges that the claims fail to meet the Written Description Requirement under 35 U.S.C. § 112, first paragraph. "The purpose of the adequate written description requirement is to ensure that the inventor had possession of the claimed subject matter at the time the application was filed. If a person of ordinary skill in the art would have understood the inventor to have been in possession of the claimed invention at the time of filing,

even if every nuance of the claims is not explicitly described in the specification, then the adequate written description requirement is met." *In re Alton,* 76 F.3d 1168, 1175, 37 U.S.P.Q.2d 1578, 1584 (Fed. Cir. 1996).

There is a strong presumption that an adequate written description of the claimed invention is present in the specification as filed (see Federal Register, Vol. 66, No. 4 (January 5, 2001) page 1105, column 1 (citing *In re Wertheim*, 541 F.2d 257, 263, 191 U.S.P.Q. 90, 97 (CCPA 1976))). In cases in which the claims are drawn to a genus, it must be determined whether the art indicates that there is substantial variation among the species within the genus of the claimed subject matter (see Revised Interim Written Description Guidelines Training Materials). The Examiner should then determine whether a representative number of species is *implicitly or explicitly* disclosed (note that actual examples are not a requirement)(see Revised Interim Written Description Guidelines Training Materials). A representative number of species depends on whether one of skill in the art would recognize that the Applicant was in possession of the necessary common attributes or features of the elements possessed by the members of the genus in view of the species disclosed or claimed (Federal Register, Vol. 66, No. 4 (January 5, 2001) page 1106, column 3).

The Examiner alleges that the genera within the claimed invention are transgenic animals of any species, and PMS2 from any animal. The Examiner alleges that the Specification "does not disclose any transgenic mammals comprising PMS2 gene of human or any other species." While the specification may not provide an actual example of such a transgenic animal, the Examiner is reminded that no actual example is required in order to satisfy the Written Description requirement. However, the Specification clearly discloses transgenic animals

including mammals (*e.g.*, cows, pigs, or goats, see page 9, line 18) which comprise a dominant negative allele of a mismatch repair gene. The specification teaches at page 8, lines 4-8 that the dominant negative allele of a mismatch repair gene may be a dominant negative form of *PMS1*, *PMS2*, *MLH1*, or *MSH2* (all of which are known to be mismatch repair genes). Thus, the Specification does disclose transgenic animals comprising dominant negative forms of mismatch repair genes, including the PMS2 gene. The Specification also specifically teaches that hPMS2-134 is an example of a dominant negative allele of a mismatch repair gene that may be used in accordance with the invention (see page 7, lines 1-12). Thus, the Examiner's assertion that the Specification "does not disclose a hypermutable transgenic animal either that comprises a protein that comprises the first 133 amino acids of PMS2" is erroneous.

The Applicants disclose a representative number of species of animals in the Examiner's definition of genus regarding "any transgenic animal or mammal." The Specification discloses at page 9, lines 15-20 wherein it is taught that the transgenic animals may be, for example, domestic livestock including cows, pigs, sheep, goats, and horses, as well as experimental animals such as mice, rats, hamsters, guinea pigs, and rabbits. One of ordinary skill in the art would not doubt that the Applicants were in possession of the necessary common attributes or features of the elements possessed by the members of the genus "transgenic animals" in view of the species disclosed or claimed. Thus, the Written Description requirement as to this claim element is satisfied.

As for the alleged genus "PMS2 from any animal," the specification discloses at page 7, lines 13-27 that mismatch repair genes can be obtained from the cells of humans, animals, yeast, bacteria or other organisms. The specification provides guidance on how dominant negative

alleles of such genes may be identified for use in the method of the invention. That is, cells with the introduced genes may be screened for defective mismatch repair. The specification further provides a specific example of how one can screen for defective mismatch repair using a β-galactosidase reporter assay. The phenotype of cells with mismatch repair defects is hypermutability. The specification clearly teaches that cells or animals with defects in mismatch repair (such as those having a dominant negative mismatch repair gene) have increased rates of spontaneous mutation due to reduced efficacy of DNA repair, and are therefore, hypermutable. Thus, the Examiner's allegation that the specification does not describe what the phenotype would be of an animal comprising a dominant negative allele of a mismatch repair gene or a truncated human PMS2 is erroneous.

The Examiner's assertion that "it is not possible to adequately describe the transgenic animals because the effects of expressing a mismatch repair gene can not be predicted because the art of making a transgenic animal is unpredictable..." is tantamount to stating that "since the art of making transgenic animals is unpredictable, the claims can never pass the Written Description Requirement." The Applicants invite the Examiner's attention to the Federal Register, Vol. 66, No. 4 (January 5, 2001) page 1107, column 2, wherein it is stated that the Examiner must:

> [e]stablish a *prima facie* case by providing reasons why a person skilled in the art at the time the application was filed would not have recognized that the inventor was in possession of the claimed invention as claimed in view of the disclosure of the application as filed. ***A general allegation of 'unpredicability in the art' is not a sufficient reason to support a rejection for lack of adequate written description.***

Thus, the Examiner may not reject claims to transgenic animals based on his view that the art of transgenics is unpredictable.

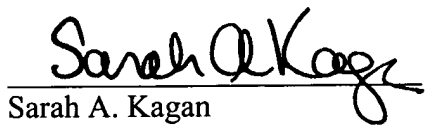
The Examiner's point regarding animals carrying truncation mutations and whether such animals would survive does not affect the Applicants' fulfillment of the Written Description requirement.

It should be noted that the claims are drawn to methods using a *dominant negative* allele of a mismatch repair gene, not wild-type, full-length PMS2, as alleged by the Examiner. The "genus" formulated in the Office Action is not as broad as alleged. The constraints on the claims include that the mismatch repair gene is a *dominant negative* allele.

It is earnestly submitted that the Application as filed reasonably conveys that the Applicants were in possession of the claimed subject matter (transgenic animals comprising a dominant negative allele of a mismatch repair gene such that the animal becomes hypermutable), and fully complies with the Written Description Requirement.

Applicants respectfully request withdrawal of the rejections and prompt allowance of the claims as amended.

Respectfully submitted,

By: 
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Version with Markings to Show Changes Made

13. (Amended) The method of claim [12] 62 wherein the mismatch repair gene is *PMS2*.

14. (Amended) The method of claim [12] 62 wherein the mismatch repair gene is human *PMS2*.

18. (Amended) The method of claim 14 wherein said mismatch repair gene [the allele] comprises a truncation mutation.

19. (Amended) The method of claim 14 wherein said mismatch repair gene [the allele] comprises a truncation mutation at codon 134 as shown in SEQ ID NO:1.

29. (Amended) The hypermutable, nonhuman, transgenic [animal] mammal of claim [28] 60 comprising a protein which consists of the first 133 amino acids of human PMS2.

52. (Amended) The hypermutable, non-human transgenic mammal of claim [50] 61 wherein the mismatch repair gene is *PMS2*.

53. (Amended) The hypermutable, non-human transgenic mammal of claim [50] 61 wherein the mismatch repair gene is human *PMS2*.

58. (Amended) The hypermutable, non-human transgenic mammal of claim [50] 61 wherein the dominant negative allele comprises a truncation mutation at codon 134 as shown in SEQ ID NO:1.

59. (Amended) The hypermutable, non-human transgenic mammal of claim 58 wherein the truncation mutation is a thymidine at nucleotide 424 of wild-type *PMS2* as shown in SEQ ID NO:1.

60. (New) A hypermutable, non-human, transgenic mammal wherein at least 50% of the cells of said mammal comprise a dominant negative allele of a mismatch repair gene.

61. (New) A hypermutable, non-human, transgenic mammal produced by a process comprising introducing a polynucleotide comprising a sequence encoding a dominant negative allele of a mismatch repair gene into said mammal, whereby said mammal becomes hypermutable.

62. (New) A method of making a hypermutable, non-human, mammalian, fertilized egg comprising introducing into said mammalian fertilized egg a polynucleotide comprising a sequence encoding a dominant negative allele of a mismatch repair gene, whereby said mammalian fertilized egg becomes hypermutable.

Clean Version of all Pending /Under Consideration Claims

13. (Amended) The method of claim 62 wherein the mismatch repair gene is *PMS2*.

14. (Amended) The method of claim 62 wherein the mismatch repair gene is human *PMS2*.

18. (Amended) The method of claim 14 wherein said mismatch repair gene comprises a truncation mutation.

19. (Amended) The method of claim 14 wherein said mismatch repair gene comprises a truncation mutation at codon 134 as shown in SEQ ID NO:1.

20. The method of claim 19 wherein the truncation mutation is a thymidine at nucleotide 424 of wild-type *PMS2* as shown in SEQ ID NO:1.

29. (Amended) The hypermutable, nonhuman, transgenic mammal of claim 60 comprising a protein which consists of the first 133 amino acids of human PMS2.

52. (Amended) The hypermutable, non-human transgenic mammal of claim 61 wherein the mismatch repair gene is *PMS2*.

53. (Amended) The hypermutable, non-human transgenic mammal of claim 61 wherein the mismatch repair gene is human *PMS2*.

58. (Amended) The hypermutable, non-human transgenic mammal of claim 61 wherein the dominant negative allele comprises a truncation mutation at codon 134 as shown in SEQ ID NO:1.

59. (Amended) The hypermutable, non-human transgenic mammal of claim 58 wherein the truncation mutation is a thymidine at nucleotide 424 of wild-type *PMS2* as shown in SEQ ID NO:1.

60. (New) A hypermutable, non-human, transgenic mammal wherein at least 50% of the cells of said mammal comprise a dominant negative allele of a mismatch repair gene.

61. (New) A hypermutable, non-human, transgenic mammal produced by a process comprising introducing a polynucleotide comprising a sequence encoding a dominant negative allele of a mismatch repair gene into said mammal, whereby said mammal becomes hypermutable.

62. (New) A method of making a hypermutable, non-human, mammalian, fertilized egg comprising introducing into said mammalian fertilized egg a polynucleotide comprising a

sequence encoding a dominant negative allele of a mismatch repair gene, whereby said mammalian fertilized egg becomes hypermutable.